

# Flowering and seed yield of lesquerella as affected by nitrogen fertilization and seeding rate

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## Abstract

The lesquerolic acid in lesquerella seed can be used in industrial applications such as greases, cosmetics, polishes, inks, and coatings. Successful commercialization of lesquerella will depend on the development of improved cultural practices. Lesquerella flowers are bright yellow and are prominently displayed. As a result, many cultural practices could be tied to flowering in a qualitative way. A new method of estimating flowers such as those of lesquerella using digital images has been developed that is rapid, not labor intensive, and can be automated. The purpose of this study was to determine the effects of nitrogen fertilizer and plant density on flowering of lesquerella and to develop relationships between seed yield and flowering. The lesquerella crop was planted on 15 October 1997, at the Maricopa Agricultural Center, approximately 40 km south of Phoenix, Arizona on a variable Mohall sandy loam (fine-loamy, mixed hyperthermic, Typic Haplargid). The experimental design was a complete factorial of three fertilizer rates and four seeding rates. Ammonium sulfate at rates of 0, 60 and 120 kg of N ha<sup>-1</sup> was applied at flowering on 18 March 1998. Digital images of the plots were taken periodically from 19 March 1998 to 4 June 1998 using a color digital camera. Images were acquired between 1030 and 1300 h MST. In this experiment, the crop did not respond to seeding rate. Flowers present at initial bloom could be used to estimate stand establishment. The early flowers did not contribute much to final yield, but flowers present in the first 3 weeks of May were a good predictor of yield. Flowering increased with N additions and noticeable peaks in flowering occurred after irrigations. The new method was verified as a viable method for estimating flower number. The method of flower estimation should also be useful for plant breeders for selection of earlier maturing lines, which would increase the potential for use of lesquerella in rotational systems. © 2003 Elsevier Science B.V. All rights reserved.

**Keywords:** Automated flower counting; Digital images; *Lesquerella fendleri*

## 1. Introduction

Lesquerella (*Lesquerella fendleri* (Gray) Wats), a newly domesticated crop, has been under study for a number of years as a source of hydroxy fatty acid (Thompson, 1988). The lesquerolic acid in

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lesquerella seed can be used in industrial applications such as greases, motor oils, cosmetics, polishes, inks, and coatings (USDA-CSRS, 1991). The current source of hydroxy fatty acids in US industry for these uses is imported castor oil (Smith, 1988).

For the desert southwest of the United States, where lesquerella has been developed, the optimum planting date is September or October (Nelson et al., 1996). Regardless of planting date, harvest dates are generally in late June (Coates, 1996; Brahim et al., 1996). Delaying harvest by 2 weeks increased potential yields (Coates, 1996), but the risk of shattering losses due to adverse weather also increased. In addition to planting and harvesting dates, lesquerella responds to plant density and fertilization (Brahim et al., 1996; Nelson et al., 1996; Brahim et al., 1998). Yields continued to increase with N applications of up to 120 kg ha<sup>-1</sup> at the beginning of flowering (Nelson et al., 1996). Brahim et al. (1996, 1998) showed populations between 750 000 and 1 000 000 plants ha<sup>-1</sup> were optimum and shattering losses increased for plant densities above 1 000 000 plants ha<sup>-1</sup>.

Water management is an important factor in lesquerella seed production. Total evapotranspiration of lesquerella was 634 mm per year of water at Maricopa, Arizona (Hunsaker et al., 1998). Yields increased linearly with water application. Water stress during peak flowering and seed development reduced yields.

In lesquerella, bright yellow flowers are conspicuously displayed at the end of stems with a marked contrast between the flowers and the vegetative portion of the plant. As a result, timing of many cultural practices such as fertilization (Nelson et al., 1996) and irrigation (Hunsaker et al., 1998) can be referenced to flowering in a qualitative manner. Previous quantitative studies of flowering have evaluated reproductive efficiency of peanut (*Arachis hypogaea* L.) (Coffelt et al., 1989), flowering density on pollinator activity in meadowfoam (*Limnanthes alba* Hartw. Ex Benth. ssp. *alba*) (Norberg et al., 1993), response to fertilizer in pumpkin (*Cucurbita moschata* Poir.) (Swiader et al., 1994), and water stress on white clover (*Trifolium repens* L.) (Turner, 1993).

Developing quantitative data on flowering has been labor intensive, especially on a daily basis. Under field conditions, frequent trips into plot areas result in mechanical damage to the plants as a result of contact. These and other difficulties associated with counting flowers have relegated flower number data to detailed studies on small numbers of plants and as not practical for daily management of a crop. Recently, estimating flower number remotely and non-destructively with digital camera derived color-images has been developed by Adamsen et al. (2000). By this method, flower counts can be accomplished without entering the plots. The purpose of this study was to determine the effects of nitrogen fertilizer and plant density on flowering of lesquerella and to develop relationships between seed yield and flowering and to verify the new flower counting method using a larger data set.

## 2. Materials and methods

### 2.1. Plot preparation and planting

The lesquerella crop was planted on 15 October 1997 in 15.2 m × 7.3 m plots at the University of Arizona's Maricopa Agricultural Center, approximately 40 km south of Phoenix, Arizona. The soil was a variable Mohall sandy loam (fine-loamy, mixed hyperthermic, Typic Haplargid) (Nelson et al., 1996). The design of the experiment was a complete factorial consisting of three fertilizer rates and four seeding rates in a randomized complete block with four replications. Seeding rates were 3.3, 6.5, 8.9 and 12.6 kg of seed ha<sup>-1</sup>. Ammonium sulfate at rates of 0, 60 and 120 kg N ha<sup>-1</sup> was applied at flowering on 18 March 1998. All plots received 54 kg ha<sup>-1</sup> of N and 26 kg ha<sup>-1</sup> of P preplant as ammonium phosphate-sulfate.

Lesquerella seed was planted very near the surface and germination was slow. Thus, it was necessary to irrigate the plots four times to keep the surface soil moist enough to achieve adequate germination (Table 1). Plots were irrigated six times during the growing season to replenish soil moisture.

Table 1  
Irrigation dates for lesquerella for the 1997–1998 growing season

Germination <sup>a</sup> irrigations	Growing season irrigations
17 October 1997	7 January 1998
21 October 1997	18 March 1998
27 October 1997	13 April 1998
8 November 1997	29 April 1998
	15 May 1998
	29 May 1998

<sup>a</sup> Planted on 15 October 1997.

## 2.2. Crop imaging

Digital images of the plots were taken periodically from 19 March 1998 to 4 June 1998 using an auto-focusing, color digital camera with an  $f$  2.8, 5 mm lens (Model D300L, Olympus America Inc., Melville, NY). The camera's nominal field of view was 57 by 42°. The camera was positioned at a constant 1.6 m above the top of the plant canopy and the position adjusted as necessary with crop growth. The camera was extended 1 m into the plot to avoid edge effects. This was accomplished by mounting the camera to an arm attached perpendicular to a telescoping pole that could be extended to a maximum height of 6 m. Thus, the same area could be imaged each time without physical entry into the plots. When images were taken, the pole was maintained in a vertical position so the camera always had a nadir view of the plot. The area selected for imaging in each plot was chosen to be representative of the plot on 19 March 1998, the first date the images were taken. Images were acquired between 1030 and 1300 h MST. The camera's built-in flash was used for each image. A white plate with red, green, and blue strips was included at the edge of each scene to provide color balance and brightness control. Estimates of flower number in the 1 m square area in each image were made by identifying all of the pixels that were flowers (Adamsen et al., 2000). Images were cropped to show only 1 m  $\times$  1 m. This reduced the field of view to 36°. Flower pixel identification was accomplished by finding all pixels with red and green values above a threshold.

The white pixels were then excluded and the remaining pixels were accepted as flower pixels.

The imaged area of each plot was hand-harvested on 26 June 1998. A 3.0 by 13.8 m strip in each plot was harvested with a combine on 30 June 1998. Analyses of variance (ANOVA) for yield and flower data were made using SAS System Version 7, Proc GLM (SAS Institute, 1999). The values for seed yield were regressed against the percent of flower pixels determined in the digital images (Adamsen et al., 2000) using the regression function in QUATTRO PRO for WINDOWS version 8 (Corel Corp. Limited, Ottawa, Canada).

## 3. Results and discussion

### 3.1. Crop response

Emergence began 8 days after planting. Additional irrigations were made on 27 October and 8 November 1997 to complete germination and establish the crop (Table 1). Differences in plant populations between seeding rates were not apparent. The percent of area covered by flowers on 19 March 1998, 1 day after N fertilizer application, showed no consistent relationship between seeding rate and initial flowering although all of the plants visible in the images were similar in size and flowering at the time. This was unexpected, but no reasons were obvious for the lack of response to seeding rate in the initial plant population.

Flowering responded to the amount of fertilizer applied, but not to the seeding rate (Fig. 1). Peaks in flowering occurred following irrigations through March and April (Table 1, Fig. 1). As the crop approached maturity in May, flowering responded to irrigation only at the lowest nitrogen level (Fig. 1a). In plots where fertilizer was applied at flowering, flower production continued at a higher rate than in the unfertilized plots which received only preplant fertilizer (Fig. 1). Peak flowering occurred on 26 March 1998 for the 0 N treatment, but not until 16 April 1998 for both the 60 and 120 kg N ha<sup>-1</sup> treatments. Flowering peaks were less pronounced and the decline in flowering was more abrupt in the 60 and 120-kg N treatments than in the no N treatment. By 4 June

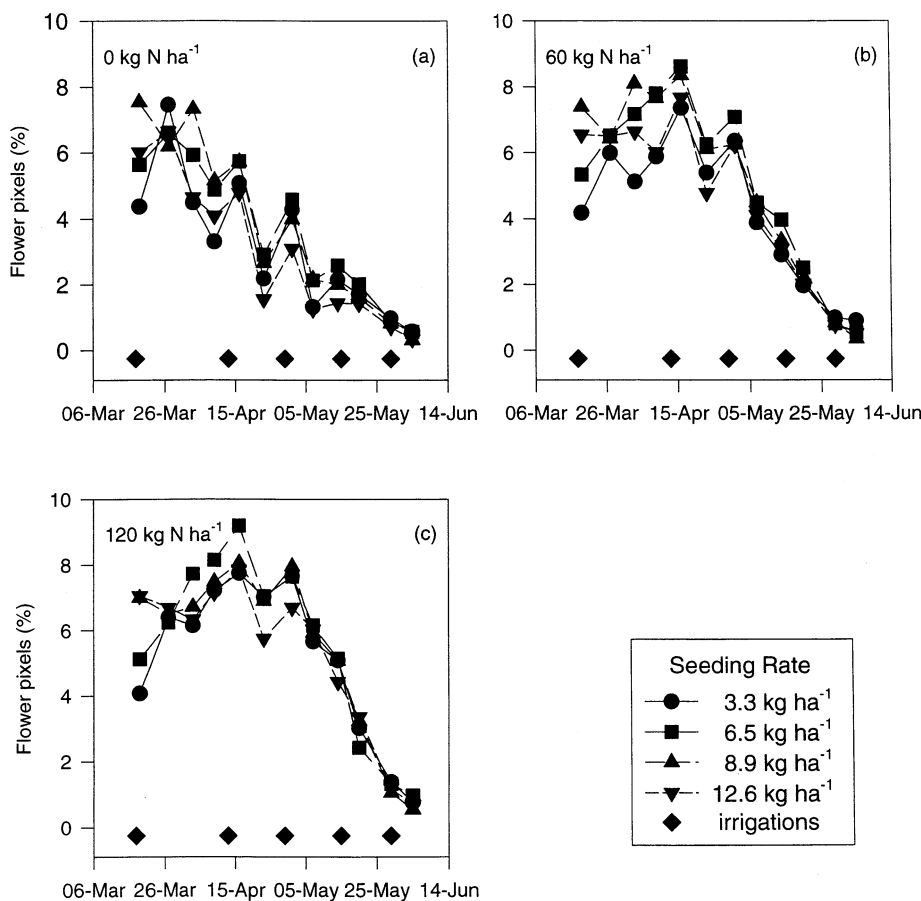


Fig. 1. Lesquerella flowering in 1998 as measured by percent flower pixels for four seeding rates and three nitrogen rates.

1998, the last date that images were acquired, all treatments had essentially stopped flowering.

Yield estimates obtained from hand harvesting the 1 m × 1 m area that had been imaged were well correlated ( $r^2 = 0.78$ ) with the yield estimates from the combine data. Combine yield estimates were 22% higher than hand-harvested values. ANOVA of both yield estimates were consistent so that only the hand-harvested data is shown (Tables 2 and 3). Yield results were consistent with the flowering data, with no significant effect of seeding rate on yield (Fig. 1 and Table 3). The seeding rate by fertilizer interaction was not significant (Table 2). Nitrogen fertilizer had a highly significant effect on yield. Each 60 kg ha<sup>-1</sup> increment of N increased seed yield by 400 kg ha<sup>-1</sup> (Table 3).

Table 2

ANOVA for lesquerella yield in 1998 from 1 m × 1 m imaged area for four seeding rates and three N levels

Source	Degrees of freedom	Mean square	F value
N level	2	2473 122	62.84*
Seeding rate	3	72 112	1.83
Reps	3	111 310	2.83
Seeding rate by N level	6	9242	0.23
Error	33	39 355	

\* Significant at 0.01 level.

Only minor differences between seeding rates were noted, and actually, the highest seeding rate was slightly lower than the two middle rates.

Table 3

Yield of lesquerella at various seeding rates and nitrogen levels in 1998 based on hand harvest of 1 m × 1 m imaged area

N level (kg ha <sup>-1</sup> )	Seeding rate (kg ha <sup>-1</sup> )				Average
	3.3	6.5	8.9	12.6	
	Yield (kg ha <sup>-1</sup> )				
0	475	561	492	366	474 c
60	760	905	939	822	856 b
120	1244	1332	1336	1127	1260 a
Average	827 AB <sup>a</sup>	933 A	922 A	772 B	

LSD<sub>0.01</sub> for seeding rate = 137. LSD<sub>0.01</sub>, for N level = 119.<sup>a</sup> Main effect means followed by the same letter in rows and columns are not different  $P = 0.01$ .

### 3.2. Digital image evaluation

Treatment means of flower pixels percentage for each date and the sum of flower pixels percentage from 19 March 1998 through each date were linearly regressed against the treatment means of yield. The coefficient of determination for the regression was then plotted against date (Fig. 2). Flowers formed in March and early April appeared to have little impact on yield. The  $r^2$  values for this period are less than 0.30, whereas the  $r^2$  values from the first 3 weeks in May were all 0.85 or higher. The largest  $r^2$  for a single date was 0.95

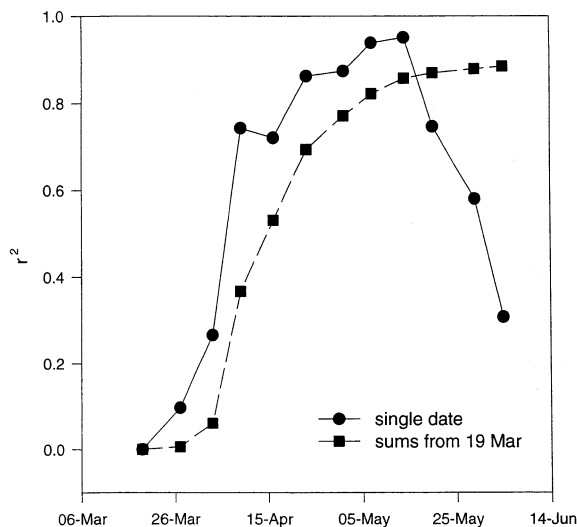


Fig. 2. Comparison of coefficients of determination ( $r^2$ ) for regressions of percent flower pixels for single dates versus yield and for the sums of percent flower pixels across dates beginning on 19 March 1998 versus yield.

for 14 May (yield =  $-3.81 + 26.03 \times \text{flower}\%$ ). For the sum of flower pixels percentage, the regression with yield never provided as good a fit as the single dates from the first 3 weeks in May. The decrease in  $r^2$  values for single date regressions after 14 May 1998 indicates that many of the flowers appearing late in the season did not mature before harvest, seeds were not formed from those flowers, or new seeds replaced old seeds lost due to shattering. Daily high temperatures in late May typically approach 40 °C and may reduce the success of pollination, making late appearing flowers ineffective.

The flowering data shows that, while flowering lasts for 12 weeks, a 4–6 week period is present beginning at 180 days after planting that has the greatest influence on yield. The number of flowers present at the beginning of flowering reflects the emergence and survival of seedlings, but the early flowers do not reflect yield. The small contribution of early season flowers to yield may have been due to a lack of pollinators early in the season; shattering of the pods formed early; the number of early flowers was small in comparison to the numbers later in the season; or a combination these or other reasons that the design of this study was unable to detect. Substantial growth occurs after fertilization at the start of flowering and most of the seeds are formed later in the growing season.

The growth of lesquerella after fertilizer application at flowering suggests that this may not be the optimum strategy for this crop. Applying fertilizer to achieve growth prior to flowering should shorten the flowering period and take

better advantage of the first flush of flowers formed by having a larger healthier plant. An impediment to early fertilizer application is the slow emergence and early growth of lesquerella. When the crop is grown with surface irrigation, as in this case, minimum water applications are 50 mm or more depending on design of the irrigation system. In this case, that means at least 200 mm of water was applied when the crop was not able to use it. Applying 200 mm of water to a fertilized crop often results in leaching of nitrate from preplant applications below the root zone (Adamsen and Rice, 1995).

Although it is not shown directly in this preliminary study, the flowering data set suggests that the reason lesquerella responds to planting date (Nelson et al., 1996) is related to growth of the plant before flowering begins. Early fall planting allows for more vegetative growth, and larger plants when flowering begins in the spring.

Bees and other pollinators are necessary for maximizing seed yields of lesquerella. Currently in large fields, growers place bees in the field when flowering begins and leave the pollinators until just before harvest. Using digital methods for monitoring flowering should enable growers to minimize the time bees are actually needed by defining when optimum flowering occurs.

The data could also be used in crop growth models associated with precision farming to make production decisions and estimate yields. Additionally, potential trouble spots can be identified. The apparent relationship between the first flower count and stand density could also provide valuable data for use in management models. Additional tests need to be done to verify these possible uses of flowering data and to quantify the relationships for different crops and environments.

#### 4. Conclusions

The ability of the flower estimation method to detect differences between plots over time also indicates that breeders could use this method to select earlier flowering lines and should lead to earlier maturity. Earlier maturity would lower water use and increase the potential use of

lesquerella in more rotational sequences. The method could also be used to develop reproductive efficiency estimates for various lesquerella populations, leading to the development of higher yielding germplasm.

Results from this study validate the method proposed by Adamsen et al. (2000) for using a digital camera to monitor flowering in a crop. They also show that by frequently monitoring flowering, critical flowering times can be identified. This can lead to altering production practices, such as earlier application of fertilizer, to maximize yield and smaller more frequent irrigations to reduce the effects of short term water stress. The cessation of flowering in conjunction with weather data should be useful in determining precise harvest dates.

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